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RECENT VIEWS ON THE NATURE OF IMMUNITY, BY ROBERT MUIR, M.D., F.R.C.P.E.

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So far as advance in medical science in the last decade of the century is concerned, the outstanding discoveries are undoubtedly those of the properties of the sera of highly immunized animals and of patients suffering from infective diseases—discoveries which have led to the establishment of sero-therapeutics and sero-diagnosis respectively. The demonstration of a definite action possessed by a serum against a toxine or a living organism in the conditions mentioned has altered former conceptions regarding the nature of immunity, and has opened up a new and wide field of research. The result has been the recognition of various properties—anti-toxic, anti-bacterial, lysogenic, agglutinative, etc., and the establishment of many interesting and important facts with regard to each. There has, however, been much dispute with regard to the exact nature of these properties and of their relation to one another, and the whole subject has become very complex. The only statement which applies to them all is that they are developed in the living body in the presence of bacteria or of separated toxines. They are all apparently the result of a reactive process; they are "antagonistic substances" in the widest sense—*Antikörper.* This statement in itself, however, suggests the question whether one fundamental process may not underlie the formation of all such anti-substances. The establishment of some general principle with regard to them is not only an important gain

from the scientific point of view, but is of much practical importance as pointing out the lines along which further advance may be more quickly and surely gained. An attempt to formulate and establish a general law has recently been made by Ehrlich in his so-called side-chain (*Seitenkette*) theory of immunity. Whether the theory will be found to be true in its entirety or not, it undoubtedly marks an epoch in the subject, and has already been the means of leading to fresh discoveries. It accordingly seems worthy to be expounded and considered.

Properties of anti-sera.—Without entering into details regarding the properties of serum in the conditions referred to, we may mention, for the sake of clearness, the chief facts. On the one hand, as is now well known, the serum of an animal treated with repeated doses of certain toxines acquires an antagonizing or neutralizing action against the particular toxine, *i.e.* is anti-toxic. Similar properties have been observed in the case of patients who have recovered from certain diseases, notably diphtheria. On the other hand, in certain conditions the serum acquires properties which are not exerted against the toxine directly, but against the bacteria: we may say, properties which induce changes in the organized bacterial structure—anti-bacterial properties. In order to the acquisition by the serum of properties of this second class the bacteria, in the living or dead condition, must themselves be introduced into the system, this being of course always the case to a greater or less extent in the natural disease. It is sufficient merely to mention with regard to these properties that the action may be *lysogenic*, leading to the dissolution and ultimate disappearance of the bacteria: or it may be *agglutinative*, leading to clumping of the bacteria; whilst, along with these, the *preventive* or *protective* property is closely associated. It is beyond our present purpose to discuss the relations of these in detail. What is of importance is that substances appear in the serum which have a definite action on the corresponding bacterium. To put these facts, with regard to anti-sera, in the most general manner, we have (*a*) in the presence of a toxine the production of a body which

neutralizes its action, and (*b*) in the presence of the bacteria the production of substances which act on bacteria of the same species. In both cases, as we shall see later, there is evidence of direct combination, *i.e.* of anti-toxine with toxine, of anti-bacterial substances with bacterial bodies. It is to be noted that in the case of all these antagonistic substances the only test we have of their existence is their physiological action. None have been obtained in a pure condition, and, though probably all are proteids of complicated constitution, even this is not known with certainty.

Relation of anti-toxine to toxine.—In considering the mode of production of antagonistic substances it is most convenient to take first the case of anti-toxines. But before doing so it will be well to consider briefly the relation of anti-toxine to toxine; in other words, how the anti-toxine acts. The two chief possibilities are (*a*) that the anti-toxine acts directly on the toxine, *i.e.* chemically, and (*b*) that the action of the anti-toxine is through the medium of the living cells, *i.e.* physiologically. It would be interesting to discuss the facts bearing on these two theories, but this has already been recently done.[1] In our opinion all the arguments go to show that anti-toxine forms a loose chemical compound with the toxine: in fact, the evidence in favour of this view seems now to be practically conclusive. On such a theory alone do the various facts with regard to the relations of the two substances receive an adequate explanation. The use of the term "chemical action" has, however, given rise to misunderstanding. This does not mean that the toxine is destroyed or broken up by the anti-toxine, merely that a chemical union has taken place. The only evidence of disappearance of the toxine is disappearance of toxic action. Now a molecule of toxine has apparently an unsatisfied atom-group by which it enters into combination with the living cells, *i.e.* has a specific affinity for part of the cell protoplasm. When toxine is combined with anti-toxine this affinity is already satisfied, and hence towards the tissues the former is rendered indifferent

[1] *Vide* Cobbett, *Journ. of Path.*, vol. vi., 1899, p. 193; also *Manual of Bacteriology*, Muir & Ritchie, 2nd ed., 1899, p. 475.

and therefore harmless—the toxic action disappears. In the treatment of a case of diphtheria, for example, the anti-toxine introduced unites, according to this view, with free toxine in the blood, and thus prevents its combining with and injuring the tissues. Certain observations brought forward by Calmette to controvert the view advanced by Fraser regarding the chemical relation of anti-venene to snake venom, and often quoted in this connection, are not really opposed to the chemical view of the relationship. In the case of snake poison the anti-toxine is less stable than the toxine, and is destroyed at a lower temperature. Now, Calmette showed that when a certain amount of toxine had been neutralized by the addition of anti-toxine, the toxic property was restored on heating the mixture at 68° C.—a temperature sufficient to destroy the anti-toxine. The objection has been recently brought forward against this result that the time element in these experiments was disregarded, a certain period being necessary before the combination is complete, and that at the time of applying heat a quantity both of free toxine and anti-toxine was present. Martin and Cherry found, on the other hand, that when sufficient time for complete combination was allowed, no toxic action reappeared on heating. But even if Calmette's interpretation is correct, the result stated does not disprove chemical union; it only proves that the toxine is not destroyed in the *physical* sense. If two complicated compounds of unequal stability are in loose chemical union, it is quite intelligible that the less stable may be destroyed (*e.g.* by heat) whilst the more stable escapes. We repeat then, that on the chemical view advocated by Ehrlich all that is implied is that an unsatisfied atom-group in the toxine becomes satisfied on uniting with the anti-toxine. As Ehrlich puts it, the anti-toxine behaves to toxine as a lock to a key All the facts with regard to the action of anti-toxine—more rapid in strong than in weak solutions and at higher temperatures than at lower, more efficient when the anti-toxine is injected with the toxine than when they are injected at different parts of the body, etc.—receive a satisfactory explanation on this theory.

In the case of anti-bacterial sera also, we have evidence that the anti-substance combines with the bacteria. This has been well shown in the case of agglutinins, these being used up in definite proportions in combining with the bacterial protoplasm or sheath. A similar statement probably applies to the lysogenic property (whether this really depends on agglutinins or not)—it certainly has been completely established in the closely analogous case of lysogenesis of red corpuscles described below. In short, to state the matter briefly, it would appear that the outstanding feature in the production of all anti-sera is the formation of bodies capable of effecting chemical union with the agent used—toxine or bacterium (or rather with some part of the latter). Having thus discussed these relations of the anti-substances, we may now consider how they are produced in the living body.

Origin of anti-toxines.—Ehrlich's theory is grounded upon his views regarding the nature of toxic action. He considers that living protoplasm may be represented as consisting of a central atom-group (*Leistungskern*) and related atom-groups or "side-chains" (*Seitenketten*). These side-chains have certain unsatisfied chemical affinities by which they fix molecules and bring them into relation to the protoplasm; they thus possess an important function in the cell economy, *e.g.* in nutrition. (The analogy is taken from the "benzol-ring" of organic chemistry.) Now there is evidence that a toxine—we are speaking of those toxines to which anti-toxines can be produced—has two essential atom-groups, one by which it becomes united to the protoplasm, and another to which the toxic effects are due. By the former it becomes anchored, as it were, to the cell; by the latter it produces a disturbance in the essential structure of the protoplasm, which is the physical basis of the toxic phenomena observed clinically. When however, the toxine is introduced in doses insufficient to produce toxic symptoms—as in anti-toxine production—a similar combination takes place with the side-chains of the protoplasm. This combination being fairly durable, the side-chains are lost for the physiological purposes of the cell, and are accordingly thrown off in combination with

the toxine molecules. A restoration of the side-chains takes place in excess of those lost, so that now more toxine can be introduced with impunity. The same result will follow as before—the combination S.-T. (side-chain + toxine) will be thrown off in greater quantity. By a continuation of gradually increasing the dose of toxine, there will ultimately be an over-production of the side-chains, *i.e.* in excess of the needs of the cell, and they become free in the blood-stream. When free they have the same chemical affinity for the toxine, and thus act as anti-toxine. As Ehrlich puts it: "The anti-toxine molecules are therefore the side-chains of the cell protoplasm regenerated in excess, and therefore discharged." The process of anti-toxine production is accordingly to be regarded as a stimulation of regeneration and over-regeneration of molecules which have been lost to the cells. Although it is stated that anti-toxine molecules are side-chains, it must be specially kept in view that they are side-chains *free in the serum.* When forming part of the protoplasm, so far from acting as anti-toxine, they really constitute the means by which the cell is rendered liable to the toxic action. On the other hand, when free in the serum they combine with the toxine, and thus prevent the latter from becoming attached to the living cells. "The same substance which, when situated in the cell, is the necessary condition of poisoning, becomes the cause of cure when it passes into the blood." (Behring.)

Such in outline is Ehrlich's theory, and we may now consider what evidence there is in support of it, and how it harmonizes with established facts. If we first of all examine the possible sources of anti-toxine, we find that they may be said to be three in number—(*a*) Anti-toxine may be formed from the toxine, *i.e.* may be a "modified toxine." (*b*) The anti-toxine may be the result of an increased formation of molecules normally present in the tissues. (*c*) The anti-toxine may be an entirely new product of living cells. It may now be considered as proved that the anti-toxine is not a modified toxine. For example, it has been shown in the case of both tetanus and diphtheria that when an animal is bled the total amount of anti-toxine in the blood may afterwards be greater

than it was immediately after the bleeding, even although no additional toxine has been introduced. This shows that the anti-toxine must be *formed* by the living cells of the body. There are other facts against this theory, which need not be detailed. Apart from the direct evidence, it is scarcely conceivable that the living body should have the power of converting the various toxines into substances which act as specific anti-toxines. It may, accordingly, be stated that anti-toxines are products of the living cells, and the third possibility mentioned—also difficult to understand on biological grounds—will be disproved if the second, as enunciated by Ehrlich, is established. In discussing the evidence in support of it, the following observations may be first of all brought forward:

(1) Ehrlich's theory offers at once an explanation of the difference between active and passive immunity, *i.e.* the immunity produced by repeated injections of toxine and that produced by injection of anti-toxine already formed. As is well known, the former is of much longer duration than the latter, a fact which is easily intelligible on the view that the cells have acquired the habit of anti-toxine formation, which is really a regenerative property. In passive immunity this property will not be brought into play at all. It would, however, be difficult to explain the difference mentioned if the anti-toxine were a modified toxine. Why, on such a supposition, should not the anti-toxine disappear from the blood as quickly in active as in passive immunity?

(2) It affords an explanation of the power which the animal body has of forming a variety of anti-toxines possessing a specific relationship to the corresponding toxines. This has always been a great difficulty in explaining the origin of anti-toxines. But if the action of a toxine depends upon a chemical affinity for certain atom-groups in the cells of the body,[1] there must be as many varieties of atom-groups as there

[1] Theoretically two different toxines might have a similar combining atom-group, and in this instance their anti-toxines would be similar. This is probably so in the case of the vegetable poisons robin and ricin (Ehrlich). But as a general rule the facts are as indicated.

are toxines, and from what has been stated above it will be evident that thus the basis for a corresponding number of anti-toxines is afforded. This power of forming anti-toxines of various kinds, which appears at first an extraordinary phenomenon, is thus to be referred back to the complicated chemical constitution of living cells. On no other theory can a rational explanation of the various specific anti-toxines be given.

(3) It should be noted that the theory implies no new process in biology; it represents merely regeneration after loss. It is in fact an application of what Weigert holds to be a general law, namely, that the bioplastic or formative processes in living tissues are always brought into play by previous loss or damage (*Schädigung*). In this connection he distinguishes a purely histo-chemical *Schädigung*, where molecules are broken down or lost without functional disturbance, from a clinical *Schädigung*, where symptoms of impaired function or of poisoning follow. The formation of anti-toxines depends upon the former: toxic action is an example of the latter. Yet the difference is rather one of extent than of kind. There is one point, however, to which further reference may be made, viz. the regeneration in excess of what has been lost, the over-regeneration implied. At first sight there may appear to be something irrational in such a supposition. But if corresponding formative processes are examined an analogy will be readily found. As Weigert points out, a similar phenomenon is probably exemplified in the case of functional hypertrophy, *e.g.* of muscle. Here also an excessive breaking down, namely, of the molecules of the contractile fibrils is the first phenomenon, and this, if continued or frequently repeated within certain limits, is not merely followed by restoration or repair, but by a repair in excess resulting in actual enlargement of the fibre. True, in this case, there is no evidence that over-regenerated molecules are discharged from the cells, yet the formative process is closely analogous to that on which Ehrlich's theory depends. So also in repair of tissues, the newly formed cells are often more numerous than those which have been lost; and, further,

this over-production or excessive proliferation becomes more marked where there is a long-continued gradual loss. As an example of this may be offered the broad zone of granulation-tissue forming the wall of a chronic sinus. In all these processes there is evidence of provision for a further and greater loss—of molecules or cells, as the case may be.

In the case of anti-toxines the amount formed strikes one as enormous. It must, however, be kept in view that the only means we have of measuring the amount is a physiological one, viz. against the action of the toxine. How much *ponderable* matter this represents we have no data for estimating. Everything, however, points to its being extremely small, just as the lethal dose of toxine is almost inconceivably minute.

If these anti-toxines are merely cell molecules which have become free in the blood stream, the question may be reasonably put—have we any evidence of the existence of such in the normal tissues? An affirmative answer has been afforded in a striking way in the case of tetanus. According to Ehrlich's theory the symptoms in this disease should be due to a combination of the toxine molecules with the side-chains of the nerve-cells. Wassermann and Takaki were the first to show that this combination actually occurs. They found that in the central nerves there are atom-groups which combine with the tetanus toxine, and therefore act as anti-toxines when tested in another animal. This was shown by bruising the spinal cord and brain of a guinea-pig so as to form an emulsion, adding a quantity of the emulsion to a certain dose of tetanus toxine and injecting into another animal. In this way they found that 1 c.c. of such emulsion could protect a mouse against ten times the lethal dose of the toxine. This, however, probably gives only a rough idea of the amount of this combining substance in the central nervous system, as the conditions after death are not the same as during life. But these results, which have been fully confirmed, demonstrate the special chemical affinity of the tetanus toxine for bodies in the central nervous system. Furthermore, emulsions of other organs tested in the same way are without effect. It may also be mentioned that a corresponding result has been

obtained by injecting the toxine in the living animal. In certain cases it has been found that after a lethal dose has been administered, free toxine can be detected in all the parts of the body except in the nervous system. In the latter situation the toxine has combined with the cells, and, as will appear from what has been said above, therefore cannot be demonstrated. Regarding the exact nature of these bodies in the nervous system practically nothing is known, but it has been found that, as regards sensitiveness to high temperatures and other physical conditions, they behave exactly as anti-toxines prepared in the usual way. It accordingly appears impossible to escape the conclusion that these atom-groups or side-chains in the normal nerve-cells and the anti-toxine molecules of tetanus are the same, *i.e.* that the latter are merely free side-chains.

In the interpretation of these results, however, there has been considerable misunderstanding on the part of various workers. The sensitiveness of the nervous system to tetanus toxine has been supposed to be a fact irreconcilable with the presence in it of anti-toxine molecules. But, according to Ehrlich's theory, the two facts are really in harmony. In the living body the molecules in question form connecting links between the toxine and the cell protoplasm: it is only when they are free in the blood stream, or, as in the above experiments, artificially brought into contact with the toxine, that they act as anti-toxines, *i.e.* as protectors of living cells. Accordingly, certain experiments of Roux and Borrel brought forward as evidence against Ehrlich's theory are really not so. They showed that the lethal dose of tetanus toxine for rabbits is much smaller when the injection is made directly into the central nervous system than in the case of subcutaneous injection. This, however, only proves that by this method combination with nerve-cells by means of their side-chains is more easily effected, and is in accordance with the theory of toxic action as above detailed. The rationale of the recently introduced method of treating tetanus by intra-cranial injection of the anti-toxine implies the same principle: the anti-toxine molecules (free side-chains) combine with any

free toxine in the nervous system and prevent its union with the side-chains of the living cells,—may probably even produce some degree of disassociation when such union has taken place.

This recent work on tetanus has accordingly yielded valuable support to Ehrlich's theory. Investigations on similar lines have also been carried out in other diseases, though the results have been much less striking. Tetanus is, however, peculiar in the definite way in which the symptoms are related to a particular tissue: in most other diseases the anti-toxines or other antagonistic substances have probably a more general origin. It was found by Wassermann in the case of typhoid, and by Pfeiffer and Marx in the case of cholera, that in the early stages of immunization the spleen, lymph-glands, and bone-marrow were especially rich in the anti-typhoid and anti-cholera substances respectively. Metchnikoff, in a recent paper, advances the view that the large hyaline leucocytes, the "macrophages," are an important source of such substances. Regarding this, however, no definite statement can yet be made. In addition to these examples in which specific combining molecules have been shown to exist in the cells of the tissues, we may also mention that traces of anti-toxines have, in certain instances, been demonstrated in the fluids of the body, *e.g.* anti-venene in the bile of the ox (Fraser), diphtheria anti-toxine in the blood serum of certain animals.

So far we have considered Ehrlich's theory as applied to the nature of anti-toxines. We have pointed out how it rests on his conception of toxic action and of the relation of anti-toxine to toxine, how it explains on biological principles the formation of anti-toxines, and how subsequently to its being brought forward it has been confirmed and supported by various recently established facts of high importance. We have now to inquire whether it can be applied to anti-bacterial sera. It is scarcely necessary to repeat that such sera have a direct action not against the toxines, only against the bacteria producing them. Nevertheless the two sera have many points in common. In addition to their being produced by an analo-

gous process, viz. the repeated injection of gradually increasing doses, the laws of passive immunity apply to both—definite substances appear in the serum of the animal actively immunized, and thus the injection of such serum into another animal confers passive immunity upon it. In both varieties of serum the potency which may be reached is very remarkable.

Lysogenesis.—As already stated, anti-bacterial sera may have various properties, but for the sake of simplicity we shall consider only one, viz. the *lysogenic*, *i.e.* the power of producing a solution or destruction of the corresponding bacterium. Pfeiffer was the first to show that if an animal were highly immunized against a particular bacterium (*e.g.* the cholera spirillum or the typhoid bacillus), and if a number of the bacteria were injected into its peritoneal cavity these underwent *lysogenesis*, *i.e.* they underwent a granular change, became swollen up and subsequently disappeared. He also showed that a similar result followed if a small quantity of the serum were injected with the bacteria into the peritoneal cavity of a fresh animal. Subsequently arrangements were devised by other observers whereby such phenomena could be produced by the anti-bacterial serum outside the body. Here also, as in the case of toxines, the action of such a serum is within certain limits specific, *i.e.* is applied only against the organism employed in its production. It should also be kept in view that this power of dissolving organisms is no absolutely new faculty, but may be possessed in small degree by normal serum towards innocuous or much attenuated organisms. In this case, however, there is no trace of specific action.

How then can such a specific dissolving property be explained on Ehrlich's side-chain theory? Theoretically the results with regard to anti-toxines might be directly applied. The action on the bacterial cell might be supposed to be due to the presence of some body in the serum which combines with it and causes disintegration of its structure. (This action is probably analogous to the destruction and disappearance of the nuclei of animal cells, *e.g.* those of the kidney under the influence of toxines.) Such bodies in the serum

would be used up in the process and would be subsequently regenerated, the regeneration gradually increasing in amount as the process is repeated. While this might theoretically be the case, it is found, however, that the process is a more complicated one than that indicated. Here not a single substance, as in the case of anti-toxines, but two substances are concerned—the action of the serum is a dual one. If such a serum is heated to 58° C. it loses its lysogenic property; it, however, regains it on the addition of a small quantity of serum from a normal animal. If the fresh serum be itself heated to 58° C. before being added it has no effect. There is, therefore, an unstable ferment-like substance present even in normal serum which is essential to the process of lysogenesis. The specific substance, that is the substance specially developed in the process of immunization, is more stable but cannot act alone; it forms as it were a link between the bacterial protoplasm and the more unstable body in normal serum. The specific substance is called by Ehrlich the immune-body (*Immunkörper*), by Bordet the "substance sensibilisatrice." When agglutination is possessed by an anti-bacterial serum it is retained when the serum is heated to the above temperature. It is still matter of dispute whether or not the agglutinin and the immune-body are one and the same, and this point need not be discussed here. We can say at least, however, that in all probability the agglutinin is a substance formed in the same way and has a combining affinity for the sheaths of the bacteria, producing in them a physical change as Gruber and Durham suppose. As a bacterium is a complicated structure from the chemical point of view, it is quite likely, according to Ehrlich's law, that more than one anti-substance is formed. In any case the side-chain theory if established will almost certainly apply to the production of agglutinins.

This remarkable lysogenic action, which may be possessed by an anti-bacterial serum, is, however, not restricted to the case of bacteria. The researches of Bordet and of Ehrlich and Morgenroth have shown that similar properties—*caeteris paribus*—can also be acquired towards red corpuscles. It has been known for some time that the blood serum of some animals

has the power of dissolving the red corpuscles of certain other animals. This, however, is not a general rule. But if an animal, whose serum has no dissolving power, is treated with repeated injections of the blood of another animal, this power becomes developed, and may reach a high degree. Bordet showed that the serum of guinea-pigs treated in this way with the blood of the rabbit, acquires powerful haemolytic properties towards the red corpuscles of the rabbit, and the same is true in the case of various other animals. The haemolytic property is, as a rule, exerted only or chiefly towards the blood of the animal employed—that is, within certain limits it is specific Sometimes the serum acquires agglutinating power towards the same red corpuscles; sometimes this power is absent. Now, the striking fact with regard to this haemolytic process is that its mechanism is practically the same as that of the lysogenesis of bacteria. In the former, just as in the latter, two substances are concerned. One is the specific substance, or, to be more accurate, the substance specially developed in excess—the "immune-body": it is the more stable, and resists heating at 58° C. The other is a ferment-like substance, the "addiment" of Ehrlich, which is present in normal serum: it is the less stable, being readily destroyed at 58° C. A serum heated to this temperature accordingly loses its haemolytic property, but regains it when a little fresh serum is added. The immune-body thus appears to form a link between the addiment and the red corpuscles—in other words, the conditions of haemolysis are practically the same as those of bacteriolysis.

The relations of the bodies in a haemolytic serum are more easily studied, and this has already been done to a fuller extent than in the case of bacteriolysis. Ehrlich and Morgenroth have proved, for example, that the immune-body enters into direct chemical union with the red corpuscles. This was shown in the following way: A quantity of haemolytic serum sufficient to dissolve a known amount of red corpuscles was taken and heated to 58° C.: the haemolytic function was, of course, thus nullified, the immune-body left in the serum being unable to act alone. This serum was then added to the red

corpuscles, and allowed to act for some time at a suitable temperature. The mixture was then centrifugalized, clear fluid and massed red corpuscles being thus separated. The clear fluid was then tested for the presence of the immune-body, by adding to it a little fresh serum, and thereafter observing its effects on red corpuscles. No haemolysis occurred, therefore the immune-body had disappeared from the fluid. Its presence in the red corpuscles, obtained by centrifugalization, was then tested for by adding to them a little fresh serum, and haemolysis was found to occur at once. In other words, the immune-body had completely combined with the red corpuscles. Other observations, along similar lines, showed that the addiment does not combine directly with the red corpuscles, but does so with the immune-body, though the combination is less firm than that of immune-body with red corpuscles. This process of haemolysis accordingly depends upon the bringing of the ferment-like body of normal serum into relation with red corpuscles by means of the specially developed immune-body. Accordingly, just as in anti-toxine production, the essential element is the production in excess of a definite substance which has a special chemical or combining affinity for the organic substance used in the injections, viz. the red corpuscles. The only difference is, that in lysogenesis the substance produced is also able to fix by another atom-group a ferment-like substance. Ehrlich, as already stated, considers that the normal function of side-chains is to bring molecules into relation to the cell for the purpose of nutrition. He further considers that if the molecule in question be small or comparatively simple, the side-chain merely combines with it: if, however, the molecule be of large size, and requires to be broken up for the purposes of the cell, the side-chain has also the power of fixing a ferment-like substance. The free side-chains constituting anti-toxines belong to the former class: those constituting lysogenic substances or lysines belong to the latter. So far as we know, no complete investigations on the combining relationships of the substances concerned, such as those detailed with regard to haemolysis, have been

carried out in the case of bacteriolysis; but there can be little doubt that corresponding facts will be established here also. The observations on haemolysis have opened up this field of research, and new facts will, no doubt, be quickly established. As an example of this may be mentioned a recently published paper of Metchnikoff, in which he shows that the animal body can acquire the power of dissolving other organized substances —*e.g.* leucocytes—that is, an anti-leucocytic serum may be obtained. And it is therefore quite probable that the lysogenesis of bacteria, so far from being an isolated phenomenon, will be found to be merely an instance of a general law. It does not seem unjustifiable to anticipate that important practical applications of these and like results will follow.

We have thus endeavoured to show how Ehrlich's side-chain theory has been applied to the various anti-sera. In many respects it is already more than a theory, although confirmation of various points and further investigation are still necessary before it can be considered to be completely established. The leading idea is that in the production of all such sera there is the development of substances which have a special combining affinity for the substance introduced—toxine, bacterium, red corpuscle, etc., as the case may be. The development of these anti-substances represents an over-regeneration and setting free of molecules normally present, which have been lost by combination with the particular substance introduced into the body. We have brought forward this account as the theory is in our opinion the first to afford a rational explanation of the phenomena concerned. To some it may appear to have chiefly a theoretical or speculative interest, but such is not the case. Apart from the importance of attaining a scientific explanation of serum therapeutics and serum diagnosis, and we may add of the spontaneous cure of disease, the knowledge of the processes concerned is of the highest value in the search for further practical results in this department.

BIBLIOGRAPHY.

The following list contains some of the more important papers on this subject. Though by no means complete, it may be found useful for purposes of reference.

Ehrlich.—"Die Wertbemessung des Diphtherieheilserums," Jena, 1897. "Ueber die Constitution des Diphtheriegiftes," *Deutsch. med. Woch.* 1898, No. 33.

Ehrlich and Morgenroth.—*Berlin. klin. Woch.*, 1899, Nos. 1 and 22.

Weigert, *Deutsch. med. Woch.*, 1896, No. 40. Article, "Antitoxinimmunität" in Lubarsch and Oestertag's *Ergebnisse der allgemeinen Pathologie*, etc., 1899.

Behring.—*Deutsch. med. Woch.*, 1898, No. 5; 1899, No. 1.

Roux and Borrel, *Annal. de l'Inst. Pasteur*, 1898, p. 225.

Bordet, *ibid.*, 1898, p. 688; 1899, p. 273.

Metchnikoff, *ibid.*, 1899, p. 737.

C. J. Martin and Cherry, *Proc. Roy. Soc. Lond.*, 1898, p. 420.

Wassermann and Takaki, *Berlin. klin. Woch.*, 1898, No. 4.

Wassermann, *ibid.*, 1898, p. 209.

Pfeiffer and Marx, *Zeitsch. f. Hyg.*, vol. 27, p. 272.

Plimmer, *Journ. of Path. and Bacter.*, vol. 5, p. 489.

Cobbett, *ibid.*, vol. 6, p. 193.

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